

## CLAIMS

1. A method of analyzing the spatial distribution of at least one chemical substance retained by a biological matter, characterized by the steps of
- 5 (a) supplying a sample of said biological matter as a specimen surface;
- (b) producing at least one imprint of said specimen surface on at least one corresponding separate substrate surface,
- 10 said at least one chemical substance being transferred to the same with retained lateral distribution thereon;
- (c) subjecting said at least one imprint to imaging mass spectrometry, at least one signal from at least two points being produced, the magnitude of said at least one signal
- 15 being dependent on the amount of said at least one chemical substance laterally present on said substrate surface;
- (d) recording said at least one signal from said at least two points; and
- (e) determining said spatial distribution of said at least
- 20 one chemical substance from said at least one image of said at least one imprint.
2. The method as in claim 1, wherein said at least one chemical substance mainly comprises organic material.
3. The method as in claim 2, wherein said organic
- 25 material comprises a lipid, an amino acid, a peptide, a protein, a carbohydrate, a nucleotide, a transmitter substance, a drug, or a targeting molecule.
4. The method as in claim 3 and 4, wherein said nucleotide is a DNA-molecule.
- 30 5. The method as in claim 3, wherein said targeting molecule is a complementary DNA-sequence.
6. The method as in any of claims 1-3, wherein said targeting molecule is an antibody or a fragment thereof.
7. The method as in claim 3, wherein said targeting
- 35 molecule comprises a chemical label.

8. The method as in claim 7, wherein said chemical label is an unusual element or an isotope.

9. The method as in any of claims 1-8, wherein said biological matter comprises cells, tissue, virus, body  
5 liquid, or biological molecules.

10. The method as in any of claims 1-9, wherein said sample of said biological matter is supplied as a specimen surface *in situ*.

11. The method as in any of claims 1-9, wherein said  
10 sample of said biological matter is supplied as a specimen surface by applying it on a solid surface.

12. The method as in claim 11, wherein said solid surface is a glass surface.

13. The method as in any of claims 1-12, wherein  
15 multiple sequential imprints are produced from the same area of said specimen surface.

14. The method as in any of claims 1-13, wherein said biological matter is fractured or cut in order to expose its interior before producing said at least one imprint.

20 15. The method as in any of claims 1-14, wherein said specimen surface is pretreated immediately before producing said at least one imprint.

16. The method as in claim 15, wherein said specimen surface is pretreated by condensing a liquid of a non-polar  
25 solvent and/or a polar solvent onto the same.

17. The method as in claim 16, wherein said polar solvent is a water solution.

18. The method as in claim 16 or 17, wherein said specimen surface is first brought to room temperature or  
30 cooled and is then arranged above a heated container containing said liquid.

19. The method as in any of claims 1-18, wherein said at least one imprint is produced within 100 s after said pretreatment of said specimen surface.

20. The method as in any of claims 1-19, wherein said specimen and/or said substrate is flexible.

21. The method as in any of claims 1-20, wherein said substrate surface is a metal surface.

5 22. The method as in claim 21, wherein said metal is silver, gold, palladium, platinum, nickel, chromium, or copper, preferably silver.

23. The method as in any of claims 1-22, wherein said substrate surface is structured.

10 24. The method as in claim 23, wherein said substrate surface is structured with protrusions of 0.01-5  $\mu\text{m}$ .

25. The method as in any of claims 1-22, wherein said substrate surface is polished.

15 26. The method as in any of claims 1-25, wherein said substrate surface is cleaned immediately before producing said at least one imprint.

27. The method as in claim 26, wherein said substrate surface is cleaned by means of chemical etching, plasma cleaning, or UV/ozone treatment, or a combination thereof.

20 28. The method as in any of claims 1-27, wherein said specimen surface is subjected to lyophilization, freeze-substitution, or air drying before producing said at least one imprint.

25 29. The method as in any of claims 1-29, wherein said biological matter is subjected to a salt solution before and/or after supplying said sample of biological matter as a specimen surface.

30 30. The method as is claim 29, wherein said salt is a sodium salt, a potassium salt, a copper salt or a silver salt, preferably a silver salt.

31. The method as in any of claims 1-30, wherein said at least one imprint is produced by pressing said specimen surface against said substrate surface.

35 32. The method as in claim 31, wherein said pressing is accomplished by means of a compressible material.

33. The method as in claim 31 or 32, wherein said pressing is accomplished by applying a force between 0.01 and 10 MPa.

34. The method as in any of claims 31-33, wherein  
5 said pressing is performed for up to 100 s.

35. The method as in any of claims 31-34, wherein said pressing is performed so that said at least one imprint represents below 5 monolayers, preferably below 2 monolayers, comprising said at least one chemical substance  
10 on said substrate surface.

36. The method as in any of claims 21-27, wherein a metal layer is deposited onto said substrate surface before producing said at least one imprint.

37. The method as in any of claims 1-35, wherein a  
15 metal layer is deposited onto said substrate surface after producing said at least one imprint.

38. The method as in claim 37, wherein said layer of metal has a thickness of less than 100 nm.

39. The method as in any of claims 36-38, wherein  
20 said layer of metal is a silver layer.

40. The method as in any of claims 1-39, wherein said imaging mass spectrometry is a Secondary Ion Mass Spectrometry.

41. The method as in claim 40, wherein said Secondary  
25 Ion Mass Spectrometry is Time of Flight - Secondary Ion Mass Spectrometry.

42. The method as in claim 40 or 41, wherein a focused beam of ions is produced by the primary ion source in said Secondary Ion Mass Spectrometry.

30 43. The method as in claim 42, wherein said ions are C<sub>60</sub>, Ga, In, or Au ions.

44. The method as in claim 43, wherein said Au ions are clusters of n ions,  $n \leq 10$ .

45. The method as in claim 42, wherein said focused  
35 beam has a diameter below 10  $\mu\text{m}$ , preferably below 1  $\mu\text{m}$ .

46. The method as in any of claims 1-45, wherein a light sensitive matrix is applied onto said substrate surface before producing said at least one imprint.

47. The method as in any of claims 1-45, wherein a  
5 light sensitive matrix is applied onto said substrate surface after producing said at least one imprint.

48. The method as in any of claims 1-45, wherein a light sensitive matrix is applied onto said specimen surface before producing said at least one imprint, a  
10 portion of said light sensitive matrix being transferred to the substrate surface when said at least one imprint is produced.

49. The method as in any of claims 1-35 and 46-48, wherein said imaging mass spectrometry is a Matrix Assisted  
15 Laser Desorption Ionisation.

50. The method as in claim 49, wherein the light source of said Matrix Assisted Laser Desorption Ionization comprises a focused laser beam, preferably an ultraviolet laser beam.

20 51. The method as in any of claims 1-50, wherein said at least one signal is recorded from an array of points on said substrate surface.

52. The method as in any of claims 1-51, wherein said at least one image is produced from said at least one  
25 signal, the colour or the brightness in each point of said at least one image being dependent on the magnitude of said at least one signal from the corresponding point on said substrate surface.